

IX SEMINÁRIO DE PREVENÇÃO E CONTROLE DE INFECÇÃO EM SERVIÇOS DE SAÚDE DE SANTA CATARINA

Resistência a antimicrobianos de cepas bacterianas isoladas de alimentos

Marília Miotto

**Florianópolis
2017**



Caracterização e quantificação de *Escherichia coli* de moluscos bivalves por PCR em Tempo Real



Genotypic and phenotypic characterization of *Escherichia coli* Isolated from mollusks in Brazil and United States



O estado de Santa Catarina é o maior produtor nacional de pescados, sendo destaque tanto na área da aquicultura quanto da pesca. O **cultivo de moluscos bivalves** merece destaque no litoral de Santa Catarina e, desde 1991, a produção vem crescendo e se tornando uma importante **atividade econômica**.



Santa Catarina:

- Maior produtor de ostras e mexilhões do Brasil;
- Segundo maior da América Latina;
- Movimentação financeira bruta estimada em R\$ 78.000.000,00 – 2015.
- 572 maricultores
- 12 municípios
- MAPA – NMP/100g de *E. coli*



Ostra (*Crassostrea gigas*)



Mexilhão (*Perna perna*)

Coleta das amostras

Brasil: Janeiro - Julho/2015

40 amostras: *Crassostrea gigas* e *Crassostrea rhizophorae*

60 amostras: *Perna perna*

Site 1: São Francisco do Sul;

Site 2: Balneário Barra do Sul;

Site 3: Penha;

Site 4: Balneário Camboriú;

Site 5: Bombinhas;

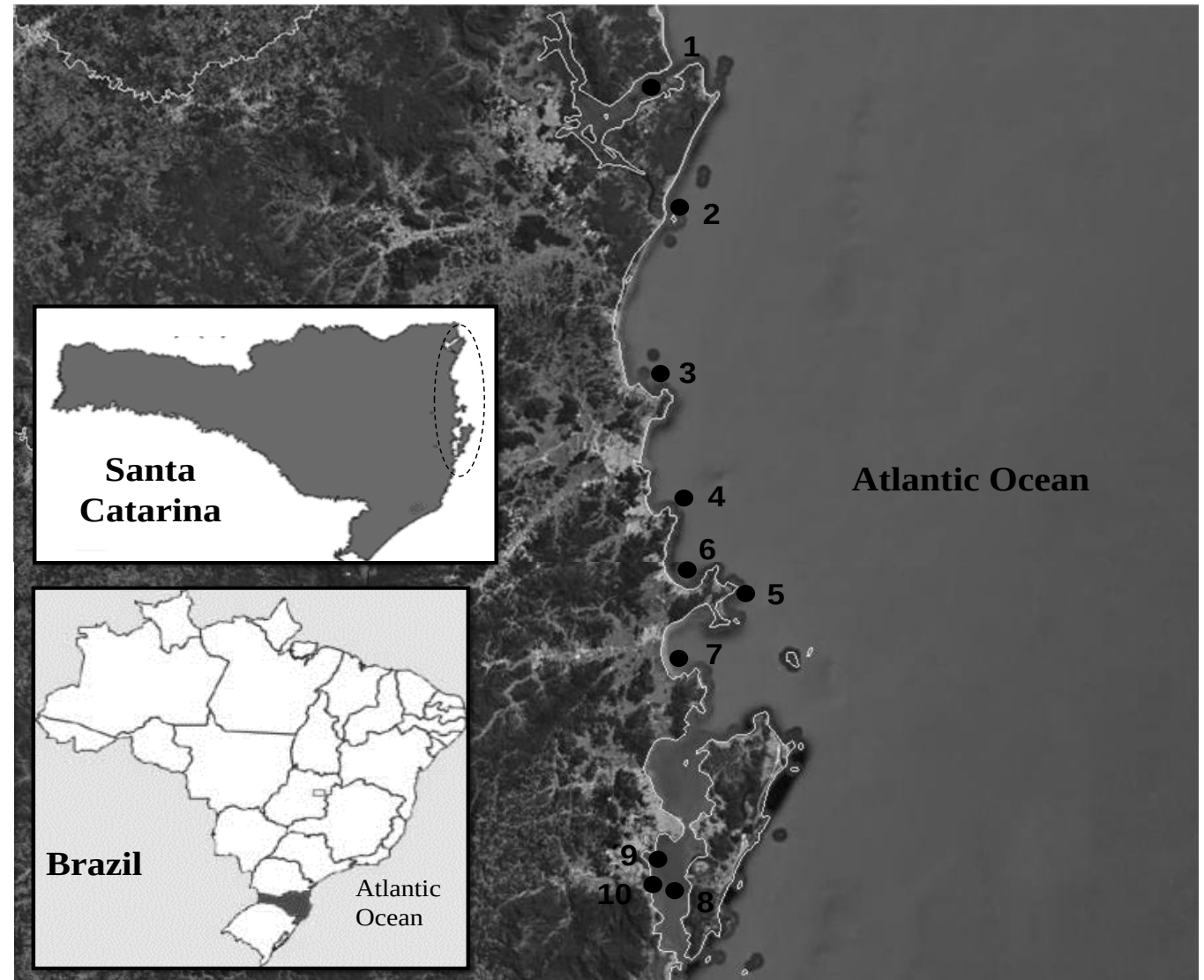
Site 6: Porto Belo;

Site 7: Gov. Celso Ramos;

Site 8: Florianópolis;

Site 9: São José;

Site 10: Palhoça.



Coleta das amostras

EUA: Outubro-Novembro/2015

18 amostras: *Crassostrea virginica*

Site A: Oxford;

Site B: Manokin;

Site C: Chester;

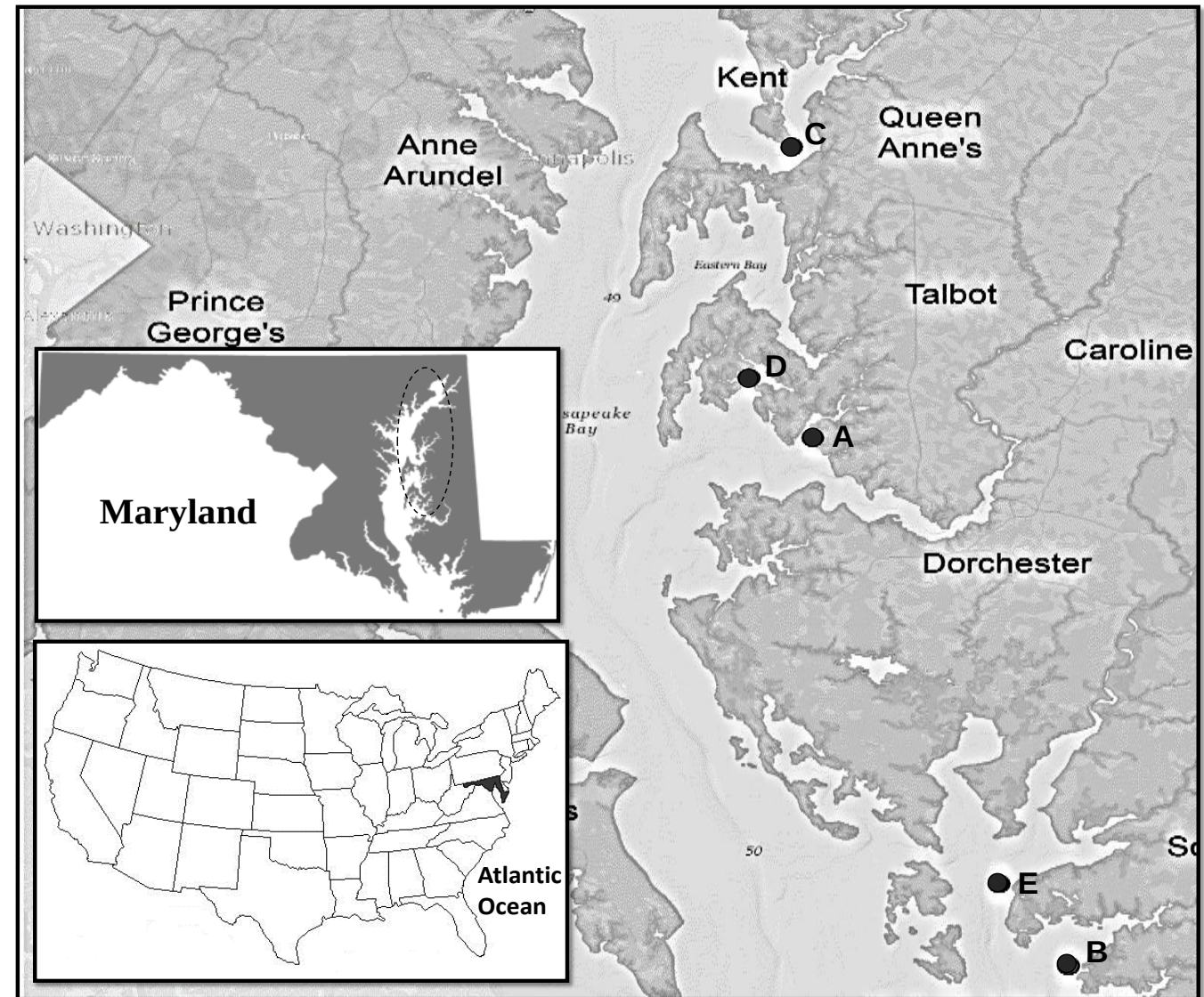
Site D: Broad Creek;

Site E: Tangier Sound

2.3 Análises microbiológicas

Método ISO 16649-3 (ISO, 2005);

NMP/100g



Teste de susceptibilidade a antimicrobianos

Método de disco difusão

Clinical and Laboratory Standards Institute (CLSI)



Antimicrobianos utilizados:

amicacina (AN, 30 μ g); amoxicillina/ácido clavulânico (AMC, 20/10 μ g); ampicillina (AM, 10 μ g); cefepime (FEP, 30 μ g); cefoxitina (FOX, 30 μ g); ceftriaxona (CRO, 30 μ g); cefalotina (KF, 30 μ g); chloranfenicol (C, 30 μ g); ciprofloxacino (CIP, 5 μ g); gentamicina (GM, 10 μ g); ácido nalidixico (NA, 30 μ g); nitrofurantoina (FM, 300 μ g); norfloxacino (NOR, 10 μ g); tetraciclina (TE, 30 μ g); trimetropina/sulfametoxazol (SXT, 1.25/23.75 μ g).

Teste de susceptibilidade a antimicrobianos

Sensível, Intermediário e Resistente: CLSI

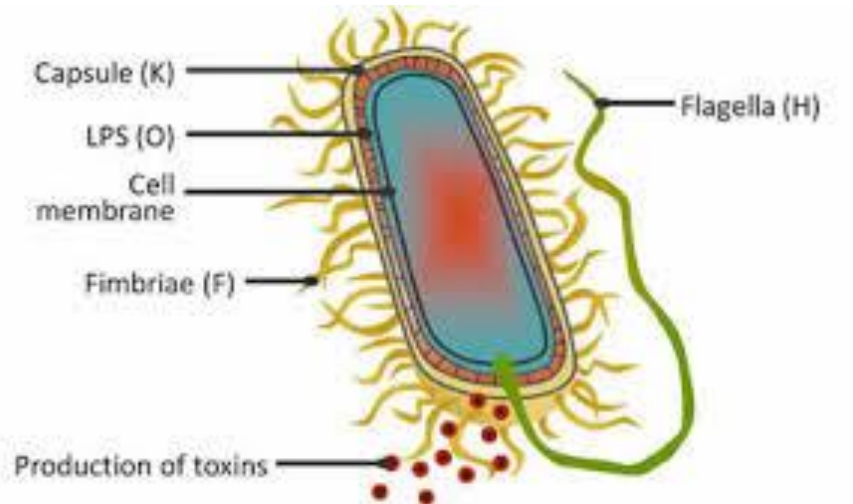
Multirresistente (MDR): resistente a três ou mais classes de antimicrobianos

Determinação dos sorogrupos

Metodologia descrita por Orskov et al., 1997;

Antisoros para os sorogrupos O1-O187 (181 antisoros);

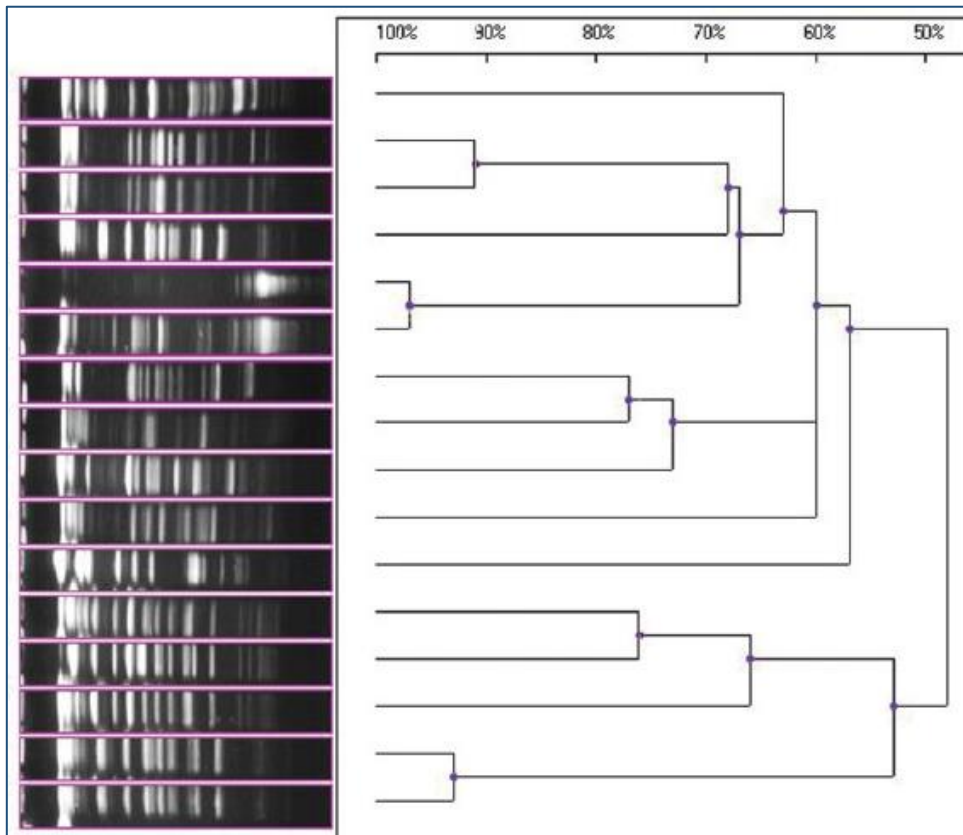
E. coli Reference Center, Pennsylvania State University, Pennsylvania, USA.



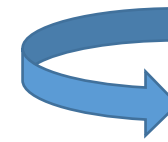
Tipagem Molecular

Bionumerics software (AppliedMaths, Austin, TX, USA);

Os perfis de PFGE foram estabelecidos com base no número e na disposição dos fragmentos e no nível de relação utilizando o coeficiente de similaridade de Dice e médias aritméticas não ponderadas (UPGMA) e coeficiente de correlação de Pearson (0,5%).

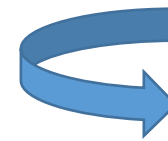


80% de similaridade



geneticamente relacionadas.

100% de similaridade



clones

Resultados

Prevalência e contagem de *E. coli*

Brasil: 100 %

20 a 18.000 NMP/100g

49% $\leq$ 230 NMP/100g

EUA: 61 %

20 a 130 NMP/100g



Brasil: 1 isolado de cada amostra positiva

EUA: 3 a 5 isolados de cada amostra positiva

141 cepas

Teste de susceptibilidade a antimicrobianos

Susceptíveis:

Cefepima (99,3 %)

Norfloxacino (97,2 %)

Cloramfenicol (95,7 %)

Trimetoprima/sulfametoxazol (91,5 %).

Resistentes:

82 % → resistente a pelo menos 1 antimicrobiano

MDR:

19,9 % → Total

26,8 % → USA

17 % → Brasil

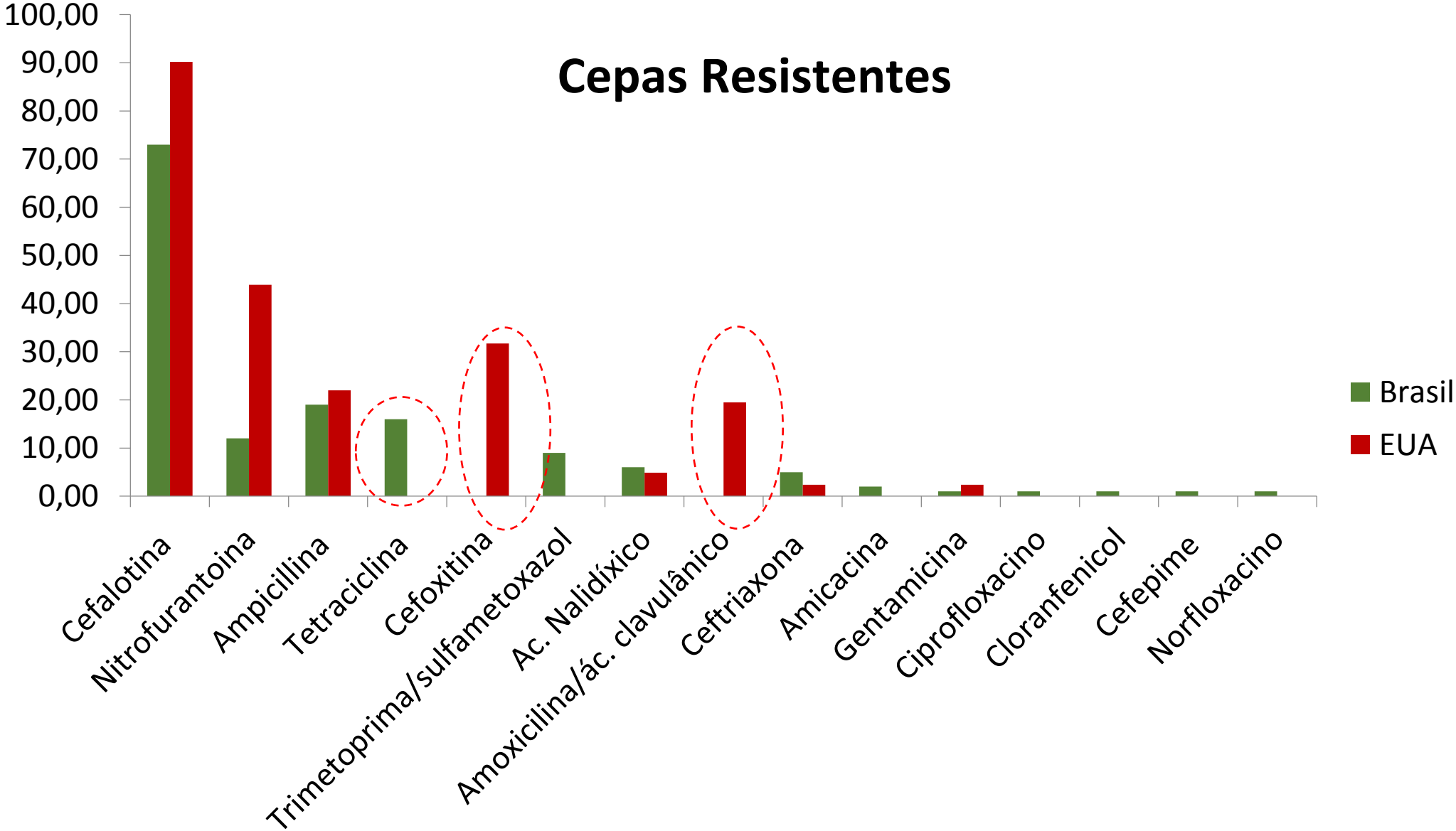


Teste de susceptibilidade a antimicrobianos

Tabela 1 – Porcentagens de isolados resistentes e com resistência intermediária aos antimicrobianos testados.

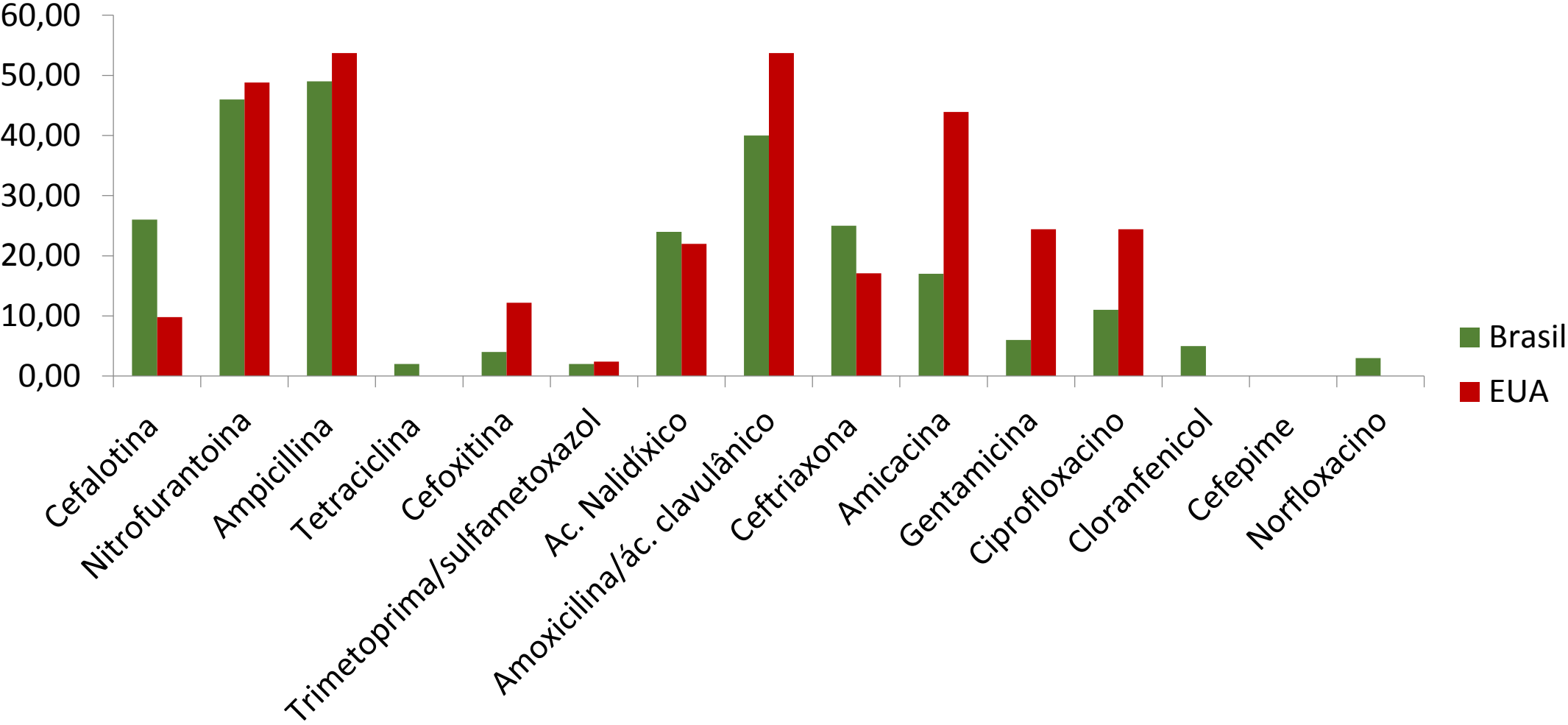
Antimicrobiano	Brasil (n = 100)		EUA (n = 41)		Total (n = 141)	
	Resistente	Intermediário	Resistente	Intermediário	Resistente	Intermediário
Cefalotina	73	26	90.2	9.8	78	22
Nitrofurantoina	12	46	43.9	48.8	21.3	46.8
Ampicillina	19	49	22	53.7	19.2	50.4
Tetraciclina	16	2	0	0	11.4	1.4
Cefoxitina	0	4	31.7	12.2	9.2	6.4
Trimetoprima/sulfametoxazol	9	2	0	2.4	6.4	2.1
Ac. nalidíxico	6	24	4.9	22	5.7	6
Amoxicilina/ác. clavulânico	0	40	19.5	53.7	5.7	44
Ceftriaxona	5	25	2.4	17.1	4.3	22.7
Amicacina	2	17	0	43.9	1.4	24.8
Gentamicina	1	6	2.4	24.4	1.4	11.3
Ciprofloxacino	1	11	0	24.4	0.7	14.9
Cloranfenicol	1	5	0	0	0.7	3.5
Cefepime	1	0	0	0	0.7	0
Norfloxacino	1	3	0	0	0.7	2.1

Teste de susceptibilidade a antimicrobianos



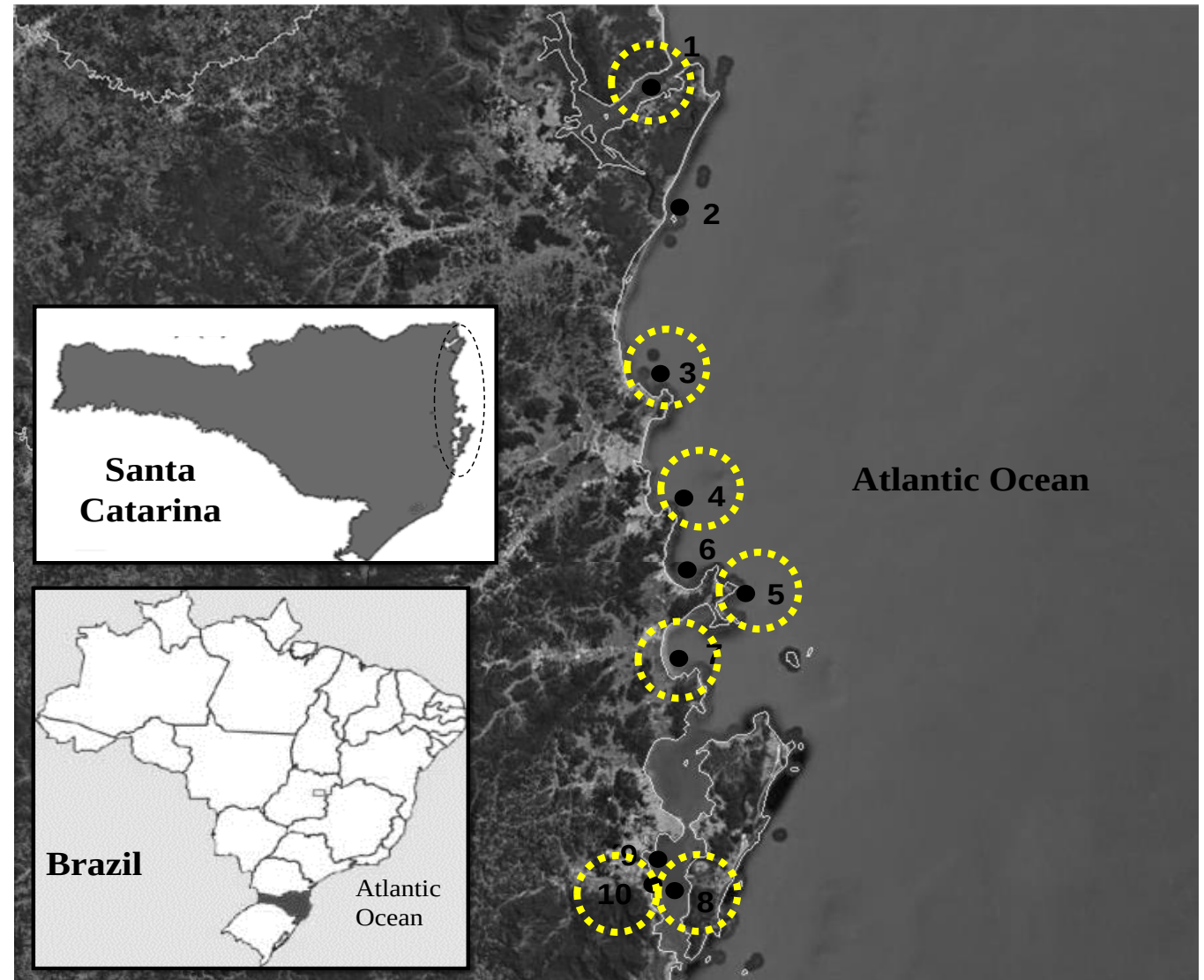
Teste de susceptibilidade a antimicrobianos

Resistência Intermediária



MDR

- Site 1: São Francisco do Sul;
- Site 2: Balneário Barra do Sul;
- Site 3: Penha;
- Site 4: Balneário Camboriú;
- Site 5: Bombinhas;
- Site 6: Porto Belo;
- Site 7: Gov. Celso Ramos;
- Site 8: Florianópolis;
- Site 9: São José;
- Site 10: Palhoça.



Determinação dos sorogrupos

O86, O126, O128 → Brasil → *E. coli* enteropatogênica (EPEC), WHO

O6, O7, O8 → *E. coli* enterotoxigênica (ETEC) → Prontos para consumo na China

O15 → Brasil → *E. coli* enteropatogênica (EPEC) → mexilhão na França, Brasil.

O5, O146 → Brasil

O113, O88 → Brasil e USA

} EPEC *E. coli* → moluscos na França

APPLIED AND ENVIRONMENTAL MICROBIOLOGY, July 1997, p. 2607–2612
0099-2240/97/\$04.00+0
Copyright © 1997, American Society for Microbiology

Vol. 63, No. 7

Association of Multiple-Antibiotic-Resistance Profiles with Point and Nonpoint Sources of *Escherichia coli* in Apalachicola Bay†

SALINA PARVEEN,^{1,2} RENDI L. MURPHREE,² LEE EDMISTON,³ CHARLES W. KASPAR,⁴
KENNETH M. PORTIER,⁵ AND MARK L. TAMPLIN^{1,2*}

Department of Food Science and Human Nutrition,¹ Department of Family, Youth and Community Sciences,²
and Department of Statistics, Institute of Food and Agricultural Sciences,⁵ University of Florida,
Gainesville, Florida 32611; Apalachicola National Estuarine Research Reserve,
Apalachicola, Florida 32320³; and Food Research Institute, University of
Wisconsin—Madison, Madison, Wisconsin 53706-1187⁴

Received 24 February 1997/Accepted 22 April 1997

A total of 765 *Escherichia coli* isolates from point and nonpoint sources were collected from the Apalachicola National Estuarine Research Reserve, and their multiple-antibiotic-resistance (MAR) profiles were determined with 10 antibiotics. *E. coli* isolates from point sources showed significantly greater resistance ($P < 0.05$) to antibiotics and higher MAR indices than isolates from nonpoint sources. Specifically, 65 different resistance patterns were observed among point source isolates, compared to 32 among nonpoint source isolates. Examples of this contrast in MAR profiles included percentages of isolates with resistance to chlortetracycline-sulfathiazole of 33.7% and to chlortetracycline-penicillin G-sulfathiazole of 14.5% for point source isolates versus 15.4 and 1.7%, respectively, for nonpoint source isolates. MAR profile homology, based on coefficient similarity, showed that isolates from point sources were markedly more diverse than isolates from nonpoint sources. Seven clusters were observed among point source isolates, with a coefficient value of approximately 1.8. In contrast, only four clusters were observed among nonpoint source isolates. Covariance matrices of data displayed six very distinct foci representing nonpoint source *E. coli* isolates. Importantly, *E. coli* isolates obtained directly from human and animal feces also clustered among point and nonpoint sources, respectively.



PRACTICE INSIGHTS

Agricultural Applications for Antimicrobials. A Danger to Human Health: An Official Position Statement of the Society of Infectious Diseases Pharmacists

Samuel L. Aitken,¹ Thomas J. Dilworth,² Emily L. Heil,³ and Michael D. Nailor,^{4,5,*}

¹Division of Pharmacy, The University of Texas MD Anderson Cancer Center, Houston, Texas; ²Department of Pharmacy, Wheaton Franciscan Healthcare – St. Francis, Milwaukee, Wisconsin; ³Department of Pharmacy, University of Maryland Medical Center, Baltimore, Maryland; ⁴Department of Pharmacy Services, Hartford Hospital, Hartford, Connecticut; ⁵Department of Pharmacy Practice, University of Connecticut, Storrs, Connecticut

The use of antibiotics in agriculture, particularly in food-producing animals, is pervasive and reported to be the largest use of antibiotics worldwide. The link between antibiotic use in agriculture and human health is unequivocal. Transmission can occur by ingestion of animal products, by direct contact of animals by animal handlers, and by environmental contamination. Antibiotics used in aquaculture and antifungals used in horticulture are also examples of antibiotic use in agriculture. Actions aimed at decreasing the amount of antibiotics used in agriculture have recently been implemented. This article describes the use of antibiotic resistance have recently been implemented to minimize antibiotic use in agriculture. This article describes t

O *link* entre o uso de antibioticos em animais e a resistencia é evidente. A transmissão pode ocorrer através da ingestão de alimentos crus que contenham bactérias resistentes, pelo contato direto com animais.... (Aitken, 2016);



Acquired Antibiotic Resistance: Are We Born with It?[▽]

Lu Zhang,^{1†} Daniel Kinkelaar,^{1‡} Ying Huang,¹ Yingli Li,^{1§} Xiaojing Li,^{1¶} and Hua H. Wang^{1,2*}

Department of Food Science and Technology¹ and Department of Microbiology,² The Ohio State University, Columbus, Ohio 43210

Received 11 April 2011/Accepted 10 July 2011

The rapid emergence of antibiotic resistance (AR) is a major public health concern. The prevalence of food-borne antibiotic-resistant (ART) commensal bacteria in ready-to-eat foods suggested that daily food consumption likely serves as a major avenue for dissemination of the food chain to human hosts. To properly assess the impact of various factors, including AR development in hosts, it is important to determine the baseline of ART bacteria in the intestinal (GI) tract. We thus examined the gut microbiota of 16 infant subjects, from the age of age, who fed on breast milk and/or infant formula during the early stages of development. Predominant bacterial populations resistant to several antibiotic resistance genes were found in the infant GI tracts within the first week of age. Several ART genes were also observed in the absence of antibiotic exposure and dietary changes. Representatives including *tet(M)*, *ermB*, *sul2*, and *bla*_{TEM} were detected in infant subjects. *Enterococcus* spp., *Klebsiella* spp., *Streptococcus* spp., and *Escherichia coli/Shigella* spp. were among the identified ART bacteria were not detected in the infant formula and infant foods examined, skin-associated ART bacteria were found in certain breast milk samples. The data suggest that development of AR in the human gut microbiota is independent of infants' exposure to antibiotics, but impacted by exposure to maternal and environmental microbes during and after delivery. The population is significantly amplified within the host even in the absence of antibiotic exposure.

The rapid emergence of antibiotic-resistant (ART) pathogens has been a critical public health concern with major social impacts. It is estimated that nosocomial infections are respon-

sibly responsible for 10% of all hospital-acquired infections, suggesting that the use of antibiotics, simply lifting the antibiotic resistance barrier, is not enough for effective control.

Após a ingestão de alimentos, a aquisição ou disseminação de genes de resistência pode ser possível através de interações com a microflora do intestino (Allen et al., 2016);

LWT - Food Science and Technology 86 (2017) 201–208



Contents lists available at ScienceDirect

LWT - Food Science and Technology

journal homepage: www.elsevier.com/locate/lwt



Antimicrobial resistance of *Lactobacillus* spp. from fermented foods and human gut

Qinqin Ma^{a,b}, Yu Fu^b, Honghu Sun^b, Yuan Huang^b, Linzhao Li^b, Qian Yu^c, Andras Dinnyes^d, Qun Sun^{b,*}

^a College of Life Sciences, Sichuan Normal University, Chengdu, Sichuan, PR China

^b Key Laboratory of Bio-resources and Eco-environment of the Ministry of Education, College of Life Sciences, Sichuan University, Chengdu, Sichuan, PR China

^c Department of Medical Technology, West China School of Public Health, Sichuan University, Chengdu, Sichuan, PR China

^d Biotalentum Ltd, Aulich u. 26, Godollo, 2100, Hungary



ARTICLE INFO

Article history:

Received 21 January 2017

Received in revised form

21 July 2017

Accepted 29 July 2017

Available online 1 August 2017

ABSTRACT

A total of 122 *Lactobacillus* strains were isolated from household-prepared or commercial yogurts and pickles in Sichuan, among which 15 representative strains, along with another 37 strains isolated from human guts, were analyzed for their antimicrobial resistance (AMR) to 17 clinically significant antimicrobials applied in China by agar dilution method. The correlation of AMRs in *Lactobacillus plantarum* from fermented foods and human gut was examined. The AMRs varied with *Lactobacillus* species. For *L. plantarum*, the distributions of minimum inhibitory concentration to 12 antimicrobials were correlated between food and gut source ($P < 0.05$), and the percentages of resistant strains from foods and gut were

Considerando que homens e animais de companhia dividem o mesmo ambiente, a transferência de bactérias resistentes ou determinantes de resistência entre animais de companhia e o homem é possível e já documentado em alguns estudos (Simjee et al., 2002; Guardabassi et al., 2004).



→ Extenso uso de antibióticos

→ Compartilhamento dos genes de resistência entre as bactérias e entre os indivíduos



Muito obrigada!!

mariliamiotto@gmail.com
miottom@saude.sc.gov.br

